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EFFECT OF DRYING TEMPERATURE IN THE ACCELERATED FREEZE-DRYING OF PORK

By A. AITKEN,* J. C. CASEY,† I. F. PENNY† and C. A. VOYLE†

The effect of plate temperature on pork *psoas* muscle dried by the Accelerated Freeze-Drying process has been studied. Plate temperatures above 60° had an adverse effect on the organoleptic and reconstitution properties of the meat. Higher plate temperatures also caused a loss in solubility of water soluble proteins and a loss in activity of phosphorylase and ATP-ase.

Introduction

A number of processes have been developed for the commercial freeze-drying of foodstuffs;[1-5] in contrast to 'classical' freeze-drying methods they supply heat to the frozen material. The processes mentioned differ mainly in the method used to supply the heat and the means of removing the water vapour. The general principles of freeze-drying of food are discussed by Harper & Tappel.[6]

No full study of the effect of process variables on the product has yet appeared. The effect of freezing rate has been examined by Wang *et al.*[7] and Rolfe.[8] Hamdy and co-workers[9] have shown that meat dried at 22–30° and 0·3–0·4 mm. Hg pressure is superior to that dried at 43° and 1·5 mm. pressure. Work at this Establishment and elsewhere has suggested that the temperature reached by the dried material may have a marked effect on the quality of the product. It was decided therefore to examine the effect of drying temperature on the properties of meat, dehydrated by the Accelerated Freeze-Drying process (AFD)[1] developed at the Aberdeen establishment. The organoleptic quality, microscopical appearance, water holding, enzymic activity, reflectance and other properties have been studied.

Experimental

Material

Pork fillets (*psoas* muscle) obtained from a local bacon factory were frozen in a commercial blast freezer to −35° and sliced across their length to give small steaks 15 mm. thick. Three steaks from each fillet were dried at each temperature. For all measurements other than by taste panel, slices were marked so that the same fillet, dried at the various temperatures, was used for a given determination. For the laboratory measurements three fillets (identified below as I, II and III) were used.

* Present address: Torry Research Station, Aberdeen
† " " Low Temperature Research Station, Cambridge

Drying process

The pork was dried by the AFD process in a small-scale vacuum dryer. The temperature of the heating plates was raised to 20, 40, 60, 80 or 100° as required and kept constant until the material was dry. The cabinet pressure, measured on a McLeod gauge, was 0·2–0·5 mm. Hg. The drying curve of the sample dried at 60° is shown as a specimen in Fig. 1.

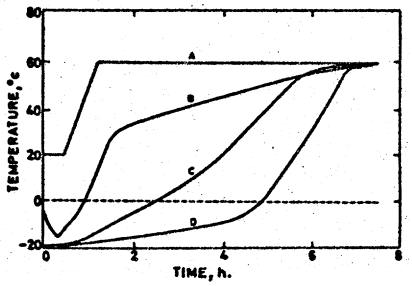

FIG. 1.—*Drying chart for pork* psoas *muscle at a temperature of 60°*

Curve A, plate temperature; B, temperature at meat surface; C and D, internal meat temperatures

pH (Fillet I)

About 1 g. of fresh or 0·3 g. of dehydrated meat was homogenised with 20 ml. of distilled water and the pH measured with a glass electrode.

Water-soluble protein

The solution prepared for phosphorylase measurements (below) was analysed by the Kjeldahl method for total nitrogen and non-protein nitrogen after trichloroacetic acid precipitation. The difference is the water-soluble protein nitrogen.

Enzymic activity

(1) *Phosphorylase*

Total phosphorylase (a and b) was determined by the method of Cori *et al.*[10] Minced dehydrated pork (0·5 g.) was homogenised with ice-cold water in a glass blender. The resulting homogenate was made up to 10 ml. and centrifuged at 5000 r.p.m. in a refrigerated centrifuge and the supernatant used for phosphorylase determinations. The final reaction mixture was $1{\cdot}4 \times 10^{-2}$M-sodium glycerophosphate of pH 6; $1{\cdot}2 \times 10^{-2}$M-cysteine hydrochloride neutralised to pH 6; 8×10^{-3}M-glucose 1-phosphate; 5×10^{-4}M-adenylic acid neutralised to pH 6; 1% glycogen and 0·08 ml. of enzyme solution to give a total volume of 0·8 ml. The glycogen, enzyme buffer and cysteine were pre-incubated at 30° for 20 min. before the glucose 1-phosphate and adenylic acid were added. After incubation for 1, 5 and 10 min., 0·05-ml. portions were withdrawn and pipetted into a mixture of 2 ml. of 0·5N-sulphuric acid and 1 ml. of 5% ammonium molybdate. Inorganic phosphate was determined by the method of Long.[11] Blanks in which the enzyme solution was boiled or the glucose-1-phosphate was replaced by distilled water were also determined.

(2) *Adenosine triphosphatase (ATP-ase)*

This was determined by the method described by Hunt & Matheson[12] for measuring ATP-ase in dehydrated meat and fish. Minced dehydrated pork (0·5 g.) was homogenised twice with ice-cold distilled water; the insoluble residue, recovered by centrifuging, was suspended in 50 ml. of water and used within 1 h. of preparation. The final reaction mixture was $2{\cdot}3 \times 10^{-4}$M-tris buffer pH 8·14; 6×10^{-3}M-$CaCl_2$; 6×10^{-3}M-ATP, 1 ml. of enzyme suspension and water to total volume 5 ml. After incubation for 1, 2 and 5 min. at 37°, 1-ml. portions were withdrawn, pipetted into 1·5 ml. of 5% trichloroacetic acid and centrifuged to

remove precipitated protein. To 1-ml. portions of the supernatant were added 1 ml. of N-H_2SO_4 and 1 ml. of 5% ammonium molybdate. Inorganic phosphate was determined by the method of Long.[11]

Glucose estimation

Glucose was extracted from the pork by blending 0·5 g. of dehydrated material with 3 ml. of 2N-perchloric acid and 3 ml. of water (or 1·5 g. of non-dehydrated pork with 3 ml. of 2N-perchloric acid and 2 ml. of water). After removal of the precipitated protein by centrifuging, 2 ml. of the supernatant were neutralised to pH 6 with 2N-potassium hydroxide and made up to 15 ml. The solution was then chilled to precipitate the potassium perchlorate which was removed by filtration. The filtrate was diluted to three times its volume and glucose determined on 1-ml. portions by a modification[13] of the glucose oxidase method of Saifer & Gerstenfeld.[14]

Reflectance

The reflectance of samples of dehydrated pork was measured in the range 370–600 mμ on a Unicam SP-500 fitted with a reflectance attachment. Measurements in this range show changes occurring in oxymyoglobin[15] and also those resulting from browning.

Contractility

Muscle fibres were teased out from the dehydrated meat and reconstituted on a microscope slide with 1 drop of 0·05M-acetate–veronal buffer pH 7·4. One drop of 0·01M-ATP was then added in close proximity to the fibre and the contraction observed with a low-power microscope.

Water-holding capacity (Fillet II)

The method is a modification of a centrifuge method used by Wierbicki *et al.*[16] Eight small pieces of dehydrated meat (about 0·5 g.) were weighed and reconstituted separately for 1 h. in 10 times their dry weight of water. Pieces of non-dehydrated frozen meat were similarly soaked in water (1·75 times their weight) for the same period. Each piece was drained for 2 min. and weighed, then wrapped in fine nylon mesh, placed in an aluminium plug as in Fig. 2, and spun for 1 h. at 3000 *g* in an angle-head refrigerated centrifuge. The weight of each piece

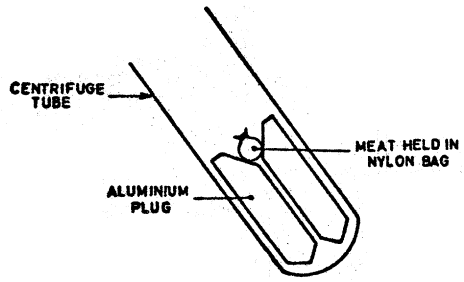

FIG. 2.—*Cross-section of aluminium plug used for centrifuging water from meat held in a nylon bag* (½ actual size)

was then measured. It had previously been shown that at 3000 *g*, the amount of loosely-bound water which was spun out of the meat became constant after 30 min. There is no clear distinction, of course, between loosely- and tightly-bound water; at higher RCF more water was spun out but did not reach a constant amount with increasing time. During the reconstitution or soaking, soluble material was leached out of the meat, altering the dry weight of the piece. The excess reconstitution (or soaking) water and the spun water were evaporated on weighed moisture dishes for 2 h. at 105° to enable corrections to be applied. The results are given as percentage moisture content, i.e., 100 × g. of water/g. of water + g. of solids (corrected).

Histological examination (Fillet III)

Materials and method.—Slices of Fillet III were dried at each of the temperatures employed as described earlier in this paper. Sections for histological examination were prepared from the dry material and also from the reconstituted material. After removal of approximately one-sixth of the dried tissue slices, the remainder was placed in distilled water at room temperature for 1 h., the depth of water being sufficient to cover the sample completely.

Longitudinal and transverse sections were prepared from portions of the dry and reconstituted material. Sections were also prepared from frozen non-dried tissue from the same source for comparison.

Reconstituted samples were placed in formol-saline for 24 h. before preparation for paraffin embedding. The dried material was placed in absolute ethanol before being embedded in the same way. The frozen non-dehydrated tissue was allowed to thaw in formol-saline at room temperature and then processed for paraffin embedding.

Sections were cut at 5μ thickness and stained to show muscle fibres and collagen fibres distinctively (Van Gieson stain). These were examined microscopically.

Taste-panel assessments

A trained panel of five or six tasters assessed the meat according to the score sheet in Table I.

Table I

Score sheet used by tasting panel for assessing the attributes of the cooked meat

Attribute	Score 6	Score 1
Odour		
Natural cooked meat	very strong	absent or masked
Processed or roast	absent	very strong
Other	absent	very strong
Flavour		
Natural cooked meat	very strong	absent or masked
Processed or roast	absent	very strong
Rancid	absent	very strong
Other	absent	very strong
Texture		
Immediate juiciness	very juicy	very dry
Immediate tenderness	very tender	very tough
Ultimate juiciness	very juicy	very dry
Ultimate tenderness	very tender	very tough

In this table, immediate juiciness and tenderness refer to the first one or two bites; ultimate juiciness and tenderness refer to these properties after thorough mastication. The arrangement of samples (unknown to the tasters) was such that each was tasted four times on different days.

Results

Drying times

The drying times shown in Fig. 3 indicate, as was anticipated, that increasing temperatures reduce the drying times from 14 h. at 20° to 5 h. at 100°.

pH

The pH values (Table II) show that a significant change in pH occurred only in the sample dried at 100°.

Water-soluble protein

Table II also shows that the solubility of the sarcoplasmic proteins falls with an increase of temperature, most rapidly at the highest temperatures.

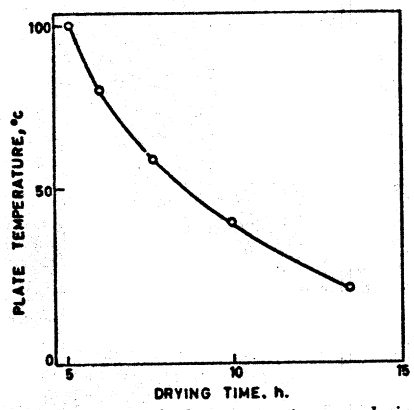

FIG. 3.—*Effect of plate temperature on drying time of pork* psoas *muscle*

Glucose

From Table II also it is seen that the amount of glucose in the dehydrated pork decreases as the plate temperature increases. A loss of glucose was also found between the non-dehydrated frozen sample and the sample dried at 20°.

Table II

Effect of drying temperature on pH, water-soluble protein and glucose content of AFD pork psoas *muscle*

Drying temperature, °C	pH	Water-soluble protein, mg. of N/g. of solids not fat	Glucose, mg./g. of solids not fat
not dried	6·18	22·8	13·80
20	6·08	22·1	9·72
40	6·10	20·3	9·68
60	6·15	20·6	8·76
80	6·07	17·0	4·32
100	5·86	11·6	1·14

Reflectance

The reflectance readings in Fig. 4 show the values in the region of 370–400 mμ which measure browning. This indicates that browning appears at drying temperatures of 80° and 100°. Visually, the samples dried at 20–60° had a pink colour and those at 80° and 100° had a definite brown colour. The results are also shown (Fig. 5) for the oxymyoglobin doublet which is quite definite at temperatures of 20–60° but which disappears at 80° and 100°. In both cases the spectrum of the 40°-sample has been omitted to avoid complicating the diagram, but it was similar to those for 60° and 20°.

Enzymic activity

Phosphorylase.—The results in Table III are expressed in terms of μg. of inorganic phosphorus produced per min. per mg. of protein N in the aqueous extract from dehydrated meat. They show that the phosphorylase activities of meat dehydrated at 20° and 60° were about equal and at 40° it was a little lower. Above 60°, however, there was a rapid drop in the phosphorylase activity.

ATP-ase.—The results in Table III are expressed as the μg. of inorganic phosphorus produced per min. at 37° per mg. of dry weight of original pork. A drop in activity was found between 20° and 40° and another drop between 60° and 80°.

Contractility

The estimations of contractility with ATP showed that muscle fibres from meat dehydrated at 20–60° were completely contractile, although at 60° the contraction was slow, but those at 80° and 100° did not respond to ATP.

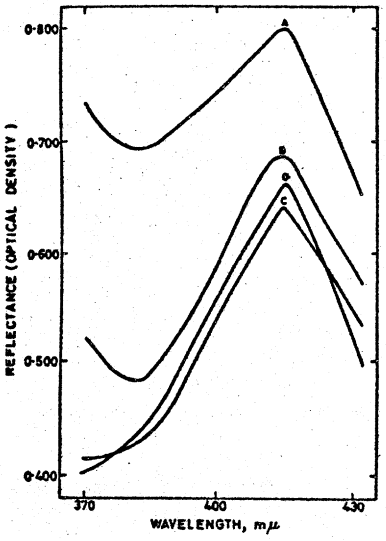

FIG. 4.—*Effect of plate temperature on reflectance of wavelengths* 370–430 *mμ of AFD pork* psoas *muscle*

Plate temperature A, 100°; B, 80° C, 60°; D, 20°

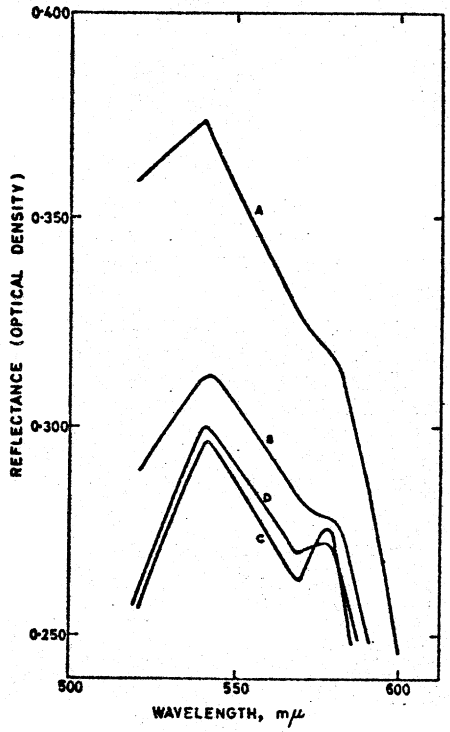

FIG. 5.—*Effect of plate temperature on reflectance at wavelengths* 500–600 *mμ of AFD pork* psoas *muscle*

Plate temperature A, 100°; B, 80°; C, 60°; D, 20°

Water-holding capacity

The moisture contents before and after spinning are plotted in Fig. 6 against temperature of drying, standard deviations also being shown. There is very little variation in the moisture contents before spinning, which is understandable as the space left by the disappearing ice

Table III

Effect of drying temperature on the activity of phosphorylase and ATP-ase of AFD pork psoas *muscle*

Drying temperature, °C	Phosphorylase, μg. of inorganic P/min./mg. of protein N	ATP-ase, μg. of inorganic P/min./mg. of solids
20	44·2	5·2
40	38·8	4·6
60	48·4	4·7
80	21·2	3·3
100	16·2	3·1

crystals is the same in all samples. The distribution of the water between fibres and interfibre space does vary, as shown (see below) by microscopical examination. The amount of loosely-held water, presumably interfibre water, which is spun out increases at a drying temperature between 80° and 100°.

The amount of soluble solids in the excess reconstitution water and the spun water decreased markedly between 80° and 100°.

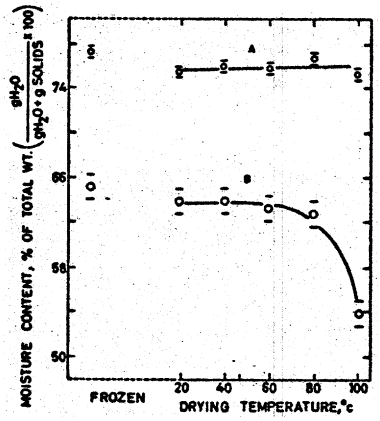

FIG. 6.—*Effect of drying temperature on water held by AFD pork* psoas *muscle after rehydration and after centrifuging at* 3000 × g *for* 5 *min.*
A, Water held after rehydration B, Water held after centrifuging

Histological examination

Sections of dehydrated material.—In each case there was the familiar pattern typical of freezing under the commercial conditions described earlier. Fairly large extra-cellular spaces separated bundles of fused fibres. There did not appear to be any differences between samples that could be attributed to variation in drying temperature.

Fig. 7 shows normal fresh *psoas* muscle in longitudinal (*a*) and transverse section (*b*) respectively. Photographs (*c*) and (*d*) show *psoas* muscle after dehydration at 60°. This may be said to be representative of all the samples, irrespective of drying temperature.

Sections of reconstituted material.—Fig. 8 (*e–j*) shows representative fields from sections of material, dried at 60°, 80° and 100°, and subsequently reconstituted. Samples dehydrated at 20° and 40° were very similar to the 60°-sample in appearance and are not illustrated here.

It would appear that reconstitution does not take place so readily in material dried at the higher temperatures (80° and 100°). Although water is taken up by these samples, it is more readily extracted again than from samples dried at lower temperatures. This water, therefore, must be absorbed into the spaces which persist between fibres, rather than into the fibres themselves. Sections (*g*) and (*i*) clearly show these spaces in reconstituted, or more accurately, rehydrated tissue, and they illustrate how loosely the water might be held between fibres.

The histological differences between samples became more marked as drying temperature was increased. No change was observed in the appearance of collagen fibres in any of the preparations. Cross-striations also were unaffected though at the magnification employed here these are not readily visible.

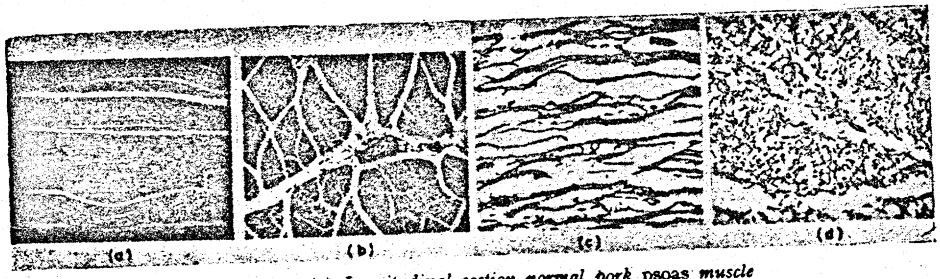

FIG. 7.—(*a*) *Longitudinal section normal pork* psoas *muscle*
(*b*) *Transverse* ,, ,, ,, ,, ,,
(*c*) *As* (*a*) *after dehydration at* 60°
(*d*) *As* (*b*) ,, ,, ,, ,,
(magnification × 15)

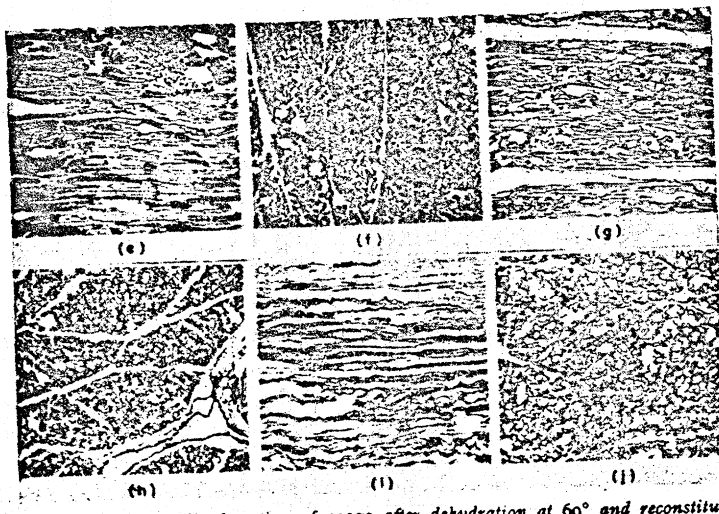

FIG. 8.—(*e*) *Longitudinal section of* psoas *after dehydration at* 60° *and reconstitution*
(*f*) *Transverse* ,, ,, ,, ,, ,, ,, ,, ,, ,, 80° ,, ,,
(*g*) *Longitudinal* ,, ,, ,, ,, ,, ,, ,, ,, ,, ,,
(*h*) *Transverse* ,, ,, ,, ,, ,, ,, ,, ,, ,, 100° ,, ,,
(*i*) *Longitudinal* ,, ,, ,, ,, ,, ,, ,, ,, ,, ,,
(*j*) *Transverse* ,, ,, ,, ,, ,, ,, ,, ,, ,, ,,
(magnification × 15)

Taste-panel assessment

The average taste-panel scores are given in Table IV. Differences in natural cooked meat odour and flavour, in processed or roast odour and flavour and in the four textural qualities are greatest between drying temperatures of 80° and 100°. These differences are statistically highly significant or very highly significant. Differences at lower temperature are not significant. These results mean that the panel was able to detect significant deterioration in quality with increasing drying temperature only above 80°.

Table IV

Taste panel assessment of pork dried at different temperatures

	Temperature of drying, °c				
	20	40	60	80	100
Odour					
Natural cooked meat	3·75	3·76	3·76	3·77	2·65
Processed or roast	4·95	5·38	4·81	4·86	3·45
Other	5·95	6·00	5·86	5·91	5·65
Flavour					
Natural cooked meat	4·05	4·14	3·81	4·23	2·70
Processed or roast	5·10	5·14	4·71	4·91	2·40
Rancid	6·00	6·00	6·00	6·00	6·00
Other	5·90	6·00	6·00	6·00	5·95
Texture					
Immediate juiciness	3·95	4·43	4·10	4·32	3·20
Immediate tenderness	3·25	3·95	3·81	3·64	2·45
Ultimate juiciness	3·60	3·91	3·72	3·82	2·30
Ultimate tenderness	3·50	3·81	4·00	3·68	2·15

Discussion

The results presented in this paper serve two purposes. First, there is the practical interest to commercial users of Accelerated Freeze-Drying (AFD). Since there is no method as yet of evaluating consumer acceptance of dehydrated meat, the taste-panel results have to be taken as a measure of the change in palatability of the product resulting from changes in temperature. It can be seen that there is no significant difference in the scores obtained for the various attributes of the meat when it was dried at any temperature between 20 and 80°. Only the meat dried at 100° showed a loss in quality. This would imply therefore that in the AFD process temperatures of up to 80° could be used without any change in the organoleptic properties being detected.

The results in this paper are also of value in indicating how far physico-chemical methods can evaluate changes which occur in meat as a result of freeze-drying.

It has been shown histologically that the higher drying temperatures, 80° and 100°, have a detrimental effect on the reconstitution of the muscle fibres. The muscle fibres did not swell to the same extent as those dried at 60° or below, and there was considerable distortion of the reconstituted fibres especially at 100°. This is confirmed by the results of water binding measured by the spinning method. Considerably more water is centrifuged out of reconstituted meat dried at 100° even though initially it absorbed the same amount of water as those dried at lower temperatures.

Glucose and browning determinations show that the subsequent storage of the dried meat may be effected by the temperature of dehydration. Visual browning appears at 80° and this is confirmed by spectrophotometric results. Some glucose however is lost even at a low drying temperature of 20° and more is lost as the temperature increases, the greatest difference being between 60–80°. This presumably implies that browning precursors, a combination of glucose with amino-groups, are formed during the drying process and, where the temperature is high, the browning proceeds more rapidly to produce visible effects.

The decrease in water-soluble protein nitrogen together with the decrease in phosphorylase activity found between 60° and 80° suggest that excessive heat treatment causes denaturation of some of the sarcoplasmic proteins. Similarly, since Hunt & Matheson[12] found that loss of ATP-ase was paralleled with loss of reconstitution properties and organoleptic quality in dehydrated meat during storage, the decrease in actomyosin ATP-ase found between 60° and 80° is indicative of changes occurring in fibrillar protein. Contractility of the muscle fibres with ATP is also lost between 60 and 80° which confirms the results obtained from ATP-ase determination.

The temperature at which meat is dehydrated by the AFD process, therefore, has a considerable effect on the properties of the resulting product. No improvement in quality is obtained by drying at temperatures below 60°, but changes occur between 60° and 80° which can be measured by physical and chemical means. Organoleptic changes can only be detected in samples heated between 80° and 100°, which suggests that taste panel assessments are less sensitive than chemical measurements. Since, however, the ultimate fate of meat is to be eaten, the changes found between 60° and 80° may be commercially insignificant.

Acknowledgments

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